1: Jpn J Cancer Res. 2000 Jun;91(6):616-21. Related Articles, Links

Japanese Cancer Association Cancer Science

Induction of cytotoxic T lymphocytes from peripheral blood of human histocompatibility antigen (HLA)-A31(+) gastric cancer patients by in vitro stimulation with antigenic peptide of signet ring cell carcinoma.

Nabeta Y, Sahara H, Suzuki K, Kondo H, Nagata M, Hirohashi Y, Sato Y, Wada Y, Sato T, Wada T, Yamashita T, Kikuchi K, Sato N.

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Antigenic peptides have been used as a cancer vaccine in melanoma patients and have led to a drastic regression of metastatic tumors. However, few antigens have been identified in non-melanoma tumors. We recently purified a new natural antigenic peptide, designated F4. 2, by biochemical elution from a human gastric signet cell carcinoma cell line and showed that it is recognized by an autologous human histocompatibility antigen (HLA)-A31-restricted cytotoxic T lymphocyte (CTL) clone. Here we describe in vitro induction of F4. 2-specific CTLs from peripheral blood T lymphocytes of HLA-A31(+) gastric cancer patients. The T cells of seven HLA-A31(+) patients with gastric cancers were stimulated in vitro by F4.2-pulsed autologous dendritic cells which had been induced from peripheral blood of each patient by incubation in the presence of granulocyte macrophage colony-stimulating factor (GM-CSF) and IL-4. We tested the cytotoxicity of the T cells against F4.2-loaded C1R-A*31012 by a 6-h (51)Cr release assay after 3 stimulations with F4.2-pulsed dendritic cells. F4.2-specific cytotoxicity was detectable in the stimulated T cells from two of the seven HLA-A31(+) patients. Further, both F4.2-specific CTLs also lysed the gastric cancer cell line, HST-2, from which F4.2 was derived. These results suggest that F4.2 peptide may be useful as an HLA-A31-restricted peptide vaccine in certain patients with gastric cancer.

PMID: 10874214 [PubMed - indexed for MEDLINE]

2: J Immunol. 1999 Sep 1;163(5):2783-91. Related Articles, Links

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Identification of natural antigenic peptides of a human gastric signet ring cell carcinoma recognized by HLA-A31-restricted cytotoxic T lymphocytes.

Suzuki K, Sahara H, Okada Y, Yasoshima T, Hirohashi Y, Nabeta Y, Hirai I, Torigoe T, Takahashi S, Matsuura A, Takahashi N, Sasaki A, Suzuki M, Hamuro J, Ikeda H, Wada Y, Hirata K, Kikuchi K, Sato N.

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Peptides of human melanomas recognized by CD8+ CTLs have been identified, but the nature of those of nonmelanoma tumors remains to be elucidated. Previously, we established a gastric signet ring cell carcinoma HST-2 and HLA-A31 (A*31012)-restricted autologous CTL clone, TcHST-2. In the present study, we determined the natural antigenic peptides of HST-2 cells. The purified

preparation of acid-extracted Ags was submitted to the peptide sequencer, and one peptide, designated F4.2 (Tyr-Ser-Trp-Met-Asp-Ile-Ser-Cys-Trp-Ile), appeared to be immunogenic. To confirm the antigenicity of F4.2 further, we constructed an expression minigene vector (pF4.2ss) coding adenovirus E3, a 19-kDa protein signal sequence plus F4.2. An introduction of pF4.2ss minigene to HST-2 and HLA-A31(+) allogeneic tumor cells clearly enhanced and induced the TcHST-2 reactivity, respectively. Furthermore, when synthetic peptides of F4.2 C-terminal-deleted peptides were pulsed to HST-2 cells, F4.2-9 (nonamers), but not F4.2-8 or F4.2-7 (octamer or heptamer, respectively), enhanced the reactivity of TcHST-2, suggesting that the N-terminal ninth Trp might be a T cell epitope. This was confirmed by lack of antigenicity when using synthetic substituted peptides as well as minigenes coding F4.2 variant peptides with Ala or Arg at the ninth position of F4.2. Meanwhile, it was indicated that the sixth position Ile was critically important for the binding to HLA-A31 molecules. Thus, our data indicate that F4.2 may work as an HLA-A31-restricted natural antigenic peptide recognized by CTLs.

PMID: 10453022 [PubMed - indexed for MEDLINE]

1: Cancer. 1995 Mar 15;75(6 Suppl):1484-9. Related Articles, Links

The mechanism of human autologous gastric signet ring cell tumor rejection by cytotoxic T lymphocytes in the possible context of HLA-A31 molecule.

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BACKGROUND. Tumor rejection antigens in human melanomas, which are recognized by cytotoxic T lymphocytes (CTLs), have recently been identified. To elucidate the cytotoxic mechanism in tumors other than melanoma, several pairs of CTLs and tumor lines were established. The authors report that HLA-A31 may present a tumor rejection antigen that is recognized by the human autologous gastric signet ring cell carcinoma-specific CTL. They also briefly describe the in vitro enhancing effect of interferon-gamma (INF-gamma) on the lysis of tumor cells by autologous CTL. METHODS. The MHC Class I-restricted CTL clone, TcHST-2, and autologous gastric signet ring cell carcinoma line, HST-2, were established. Cytotoxicity blocking assays of antibodies reacting against the MHC Class I nonpolymorphic determinant and HLA-A, B, and C haplotype elements, which are expressed on the HST-2 cells, were performed. RESULTS. Lysis of the autologous tumor cells (HST-2) by the CTL clone (TcHST-2) was enhanced when the tumor cells were pretreated with IFN-gamma. This lysis was selectively inhibited by the anti-nonpolymorphic MHC Class I determinant monoclonal antibody (MoAb) and anti-HLA-A31 haplotype-specific MoAb. However, TcHST-2 clone was not cytotoxic to HLA-A31+ allogeneic leukemia lines. CONCLUSION. Pretreatment of target cells with IFN-gamma may be a necessary procedure for the efficient lysis of HST-2 cells by autologous TcHST-2 CTL. The data indicate that TcHST-2 was MHC Class I-restricted HST-2 tumor-specific CTL and suggest that the HLA-A31 haplotype element is an antigen-presenting molecule. Also discussed is the nature of the antigenic peptides in gastric signet ring cell carcinoma.

PMID: 7889479 [PubMed - indexed for MEDLINE]

☐ **1:** J Immunother. 2003 Sep-Oct;26(5):385-93. Related Articles, Links

LWWonline

Cell transfer therapy for cancer: lessons from sequential treatments of a patient with metastatic melanoma.

Rosenberg SA, Yang JC, Robbins PF, Wunderlich JR, Hwu P, Sherry RM, Schwartzentruber DJ, Topalian SL, Restifo NP, Filie A, Chang R, Dudley ME.

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The development of effective autologous cell transfer therapies for the treatment of patients with cancer has been difficult, in part because the cells used to treat each patient are different, as are the patient's tumor and immune status. Much can thus be learned by sequential treatments of the same patient with the same cells, making single modifications in the treatments to determine which factors are critical. The authors have treated a single patient with five sequential administrations of the same cells with minor modifications in the mode of administration and the immune status of the patient. The treatment of this patient strongly suggested that 1) the highly avid recognition of tumor antigens in vitro by a transferred lymphocyte population does not necessarily predict in vivo antitumor activity; 2) the administration of highly avid antitumor autologous lymphocyte populations can be far more active in mediating tumor regression in vivo when administered after nonmyeloablative chemotherapy than when administered without this prior chemotherapy; 3) intra-arterial administration of highly avid antitumor autologous lymphocytes into the blood supply of the tumor can be more effective in mediating tumor regression than the intravenous administration of these same tumor infiltrating lymphocytes; 4) one mechanism of tumor escape from immunotherapy is loss of class I MHC antigen expression by the tumor due to mutation of the beta-2 microglobulin gene.

PMID: 12973027 [PubMed - in process]

☐ **2:** Nat Rev Cancer. 2003 Sep;3(9):666-75. Related Articles, Links

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Adoptive-cell-transfer therapy for the treatment of patients with cancer.

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Adoptive immunotherapy--the isolation of antigen-specific cells, their ex vivo expansion and activation, and subsequent autologous administration--is a promising approach to inducing antitumour immune responses. The molecular identification of tumour antigens and the ability to monitor the persistence and transport of transferred cells has provided new insights into the mechanisms of tumour immunotherapy. Recent studies have shown the effectiveness of cell-transfer therapies for the treatment of patients with selected metastatic cancers. These studies provide a

blueprint for the wider application of adoptive-cell-transfer therapy, and emphasize the requirement for in vivo persistence of the cells for therapeutic efficacy.

Publication Types:
- Review
- Review, Tutorial

PMID: 12951585 [PubMed - indexed for MEDLINE]

☐ **3:** J Immunother. 2003 Jul-Aug;26(4):332-42. Related Articles, Links

Generation of tumor-infiltrating lymphocyte cultures for use in adoptive transfer therapy for melanoma patients.

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The generation of T lymphocytes with specific reactivity against tumor antigens is a prerequisite for effective adoptive transfer therapies. Melanoma-specific lymphocyte cultures can be established from tumor infiltrating lymphocytes (TILs) by in vitro culture in high levels of IL-2. We have optimized methods for generating melanoma-reactive TIL cultures from small resected tumor specimens. We report a retrospective analysis of 860 attempted TIL cultures from 90 sequential melanoma biopsy specimens from 62 HLA-A2+ patients. Multiple independent TIL derived from a single tumor often exhibited substantial functional and phenotypic variation. Tumor specific activity was detected in TIL from 29 (81%) of 36 patients screened. TIL cultures selected for high activity were generally capable of large numerical expansion using a single round of a rapid expansion protocol. Limited clonal T-cell populations in an oligoclonal TIL culture could confer specific tumor recognition in these highly selected, highly expanded TIL cultures. These methods were efficient at generating TILs suitable for adoptive transfer therapy.

PMID: 12843795 [PubMed - in process]

☐ **4:** Proc Natl Acad Sci U S A. 2003 Jul 8;100(14):8372-7. Epub 2003 Jun 25. Related Articles, Links

Cancer regression and autoimmunity induced by cytotoxic T lymphocyte-associated antigen 4 blockade in patients with metastatic melanoma.

Phan GQ, Yang JC, Sherry RM, Hwu P, Topalian SL, Schwartzentruber DJ, Restifo NP, Haworth LR, Seipp CA, Freezer LJ, Morton KE, Mavroukakis SA, Duray PH, Steinberg SM, Allison JP, Davis TA, Rosenberg SA.

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Cytotoxic T lymphocyte-associated antigen 4 (CTLA-4) is a critical immunoregulatory molecule (expressed on activated T cells and a subset of regulatory T cells) capable of down-regulating T cell activation. Blockade of CTLA-4 has been shown in animal models to improve the effectiveness of cancer immunotherapy. We thus treated 14 patients with metastatic melanoma by using serial i.v. administration of a fully human anti-CTLA-4 antibody (MDX-010) in conjunction with s.c. vaccination with two modified HLA-A*0201-restricted peptides from the gp100 melanoma-associated antigen, gp100:209-217(210M) and gp100:280-288(288V). This blockade of CTLA-4 induced grade III/IV autoimmune manifestations in six patients (43%), including dermatitis, enterocolitis, hepatitis, and hypophysitis, and mediated objective cancer regression in three patients (21%; two complete and one partial responses). This study establishes CTLA-4 as an important

molecule regulating tolerance to "self" antigens in humans and suggests a role for CTLA-4 blockade in breaking tolerance to human cancer antigens for cancer immunotherapy.

Publication Types:
- Clinical Trial

PMID: 12826605 [PubMed - indexed for MEDLINE]

☐ **5:** Cancer J. 2001 Nov-Dec;7 Suppl 2:S51-2. Related Articles, Links

Cellular therapy: an introduction.

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Cellular therapy is defined as the transfer, to the intact host, of living cells with the intent of introducing new host functions or correcting defective functions. The appeal and the promise of cellular therapy derive from the ability to manipulate, ex vivo, cellular and molecular biologic pathways in a way that is not possible in the intact organism.

PMID: 11777264 [PubMed - indexed for MEDLINE]

☐ **6:** J Immunother. 2001 Jul-Aug;24(4):363-73. Related Articles, Links

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Adoptive transfer of cloned melanoma-reactive T lymphocytes for the treatment of patients with metastatic melanoma.

Dudley ME, Wunderlich J, Nishimura MI, Yu D, Yang JC, Topalian SL, Schwartzentruber DJ, Hwu P, Marincola FM, Sherry R, Leitman SF, Rosenberg SA.

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This report describes a phase I study of the adoptive transfer of cloned melanoma antigen-specific T lymphocytes for therapy of patients with advanced melanoma. Clones were derived from peripheral blood lymphocytes or tumor-infiltrating lymphocytes of patients who had received prior immunization with the melanoma-associated antigen, gpl00. In response to its cognate antigen, each clone used for treatment secreted large amounts of interferon-gamma and granulocyte-macrophage colony-stimulating factor, lesser amounts of interleukin (IL)-2 and tumor necrosis factor-alpha, and little or no IL-4 and IL-10. Clones also demonstrated recognition of human leukocyte antigen-matched melanomas using cytokine secretion and lysis assays. Twelve patients received 2 cycles of cells alone; 11 patients received additional cycles of cells and were randomized between two schedules of IL-2 (125,000 IU/kg subcutaneously daily for 12 days versus 720,000 IU/kg intravenously every 8 h for 4 days). A total of 51 cycles of cells were administered, with an average of 1 x 10(10) cells per cycle. Peripheral blood samples were analyzed for persistence of transferred cells by T-cell receptor-specific polymerase chain reaction. Transferred cells reached a maximum level at 1 h after transfer but rapidly declined to undetectable levels by 2 weeks. One minor response and one mixed response were observed (both in the high-dose IL-2 arm). This report demonstrates the safety and feasibility of cloned T-cell transfer as a therapy for patients with cancer. The lack of clinical effectiveness of this protocol suggests that transfer of different or additional cell types or that modulation of the recipient host environment is required for successful therapy.

Publication Types:
- Clinical Trial
- Clinical Trial, Phase I